THE ETIOLOGY OF ACUTE EPIDEMIC POLIOMYELITIS *

PLATES 1 TO 4

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The studies of the pathologic anatomy and epidemiology of acute poliomyelitis of Wickman,[1] Harbitz and Scheel,[2] and others, clearly indicated that the disease was infectious in nature, and the earlier work on its etiology was confined to the bacteriology of the infected tissues. These investigations yielded varying results. Early observers found micrococci of different types in poliomyelitis, but the experimental evidence brought forth as indicative of their etiologic importance is inconclusive. The first noteworthy advance towards proving the infectious nature of the disease was announced by Landsteiner and Popper[3] in 1909. They reported experiments in which they had transmitted the disease to 2 monkeys by the intraperitoneal injection of a saline emulsion of the spinal cord of a child that had died during the acute stage of the disease. At about the same time Flexner and Lewis[4] reported that they had transmitted the disease through several inoculation generations of monkeys. They also had produced the disease by various inoculation routes, and had demonstrated the infectious material in the spinal fluid, blood, nasopharyngeal mucosa, and lymph nodes near the site of inoculation.[5] The results of these observers were confirmed by many investigators, including Knoepfelmacher,[6] Strauss and Huntoon,[7] Leiner and von Wiesner,[8] and others. Furthermore Flexner and Lewis[9] and Landsteiner and Levaditi[10] found that the infectious agent was filterable through Berkefeld filters and thenceforth the etiologic agent has been spoken of as a filterable virus.

* Received for publication October 11, 1916.

[1] Die akute Poliomyelitis bzw. Heine-Medinische Krankheit, 1911.

[2] Pathologisch-anatomische Untersuchungen über akute Poliomyelitis und verwandte Krankheiten, 1907.

[3] Ztschr. f. Immunitäts., 1909, 2, p. 377.

[4] Jour. Am. Med. Assn., 1909, 53, pp. 1639, 1913, 2095.

[5] Jour. Exper. Med., 1910, 12, p. 227.

[6] Med. Klin., 1909, 5, p. 1671.

[7] New York Med. Jour., 1910, 91, p. 64.

[8] Wein. klin. Wchnschr., 1909, 12, p. 1698.

[9] Jour. Am. Med. Assn., 1909, 53, p. 2095.

[10] Compt. rend. Soc. de biol., 1909, 67, p. 592.

In recent years the virus of poliomyelitis has been cultivated on artificial media by Flexner and Noguchi,[11] using methods similar to those used in the cultivation of Spirochaeta pallida. In this work small pieces and emulsions of the brain and cord of monkeys dead of acute poliomyelitis were placed in tall tubes of ascites fluid containing sterile fresh rabbit kidney and the tubes incubated under anaerobic conditions. An opalescence appeared around the tissue, usually after 5 days, which increased until sedimentation occurred, about 5 days later. Examination of smears of the sediment by the Giemsa method of staining revealed the presence of small globoid bodies arranged in pairs, groups, and chains. Similar bodies were found in microscopic preparations of infected tissue and in glycerinated material. Typical lesions and death were produced in monkeys when such cultures were used for inoculation. These results led to the wide acceptance of the view that poliomyelitis is caused by a filterable virus, the bacteria that had previously been described in connection with this disease being regarded as secondary invaders.

During the summer of 1916 a severe epidemic of acute poliomyelitis prevailed in the eastern states, especially in New York, which stimulated wide-spread interest in this disease, and forcibly impressed the fact that our knowledge of its etiology and transmission was still incomplete. In Chicago a considerable number of cases occurred, from which the material for this study was obtained. In the light of accepted facts the most important problems in the etiology and transmission of acute poliomyelitis seemed to center in simplification of the methods of cultivation and identification of the infectious agent. Since it had been proved that the virus of this disease is present in the tissues of the central nervous system, fresh postmortem material was used for study; however, the blood and spinal fluid during life were studied bacteriologically in several instances. Cultures of the brain, cord, and other tissues were made on a great variety of media, some of the results of which have been announced in a previous, preliminary report.[12]

TECHNIC

The brain, spinal cord, and other material from persons dead of acute poliomyelitis were obtained under sterile conditions as soon after death as possible and cultures made immediately. Small pieces of nervous tissue were washed in sterile normal salt solution, crushed, and planted in ascites-fluid media, and ascites dextrose agar containing pieces of fresh sterile rabbit

[11] Jour. Exper. Med., 1913, 18, p. 461.
[12] Mathers, Jour. Am. Med. Assn., 1916, 67, p. 1019.

kidney, coagulated normal horse serum, and ascites dextrose broth. The cultures were incubated at 35-37 C. under both aerobic and anaerobic conditions for from 1 to 10 days. Usually growth appeared in some of the cultures in from 1 to 5 days. In the preparation of filtrates of the brains and cords sterile normal salt solution was used as a menstruum. The tissue suspensions were filtered through Berkefeld "N" filters and porcelain filters of the Maassen type. Cultures were made of the filtrates in the same media as the cultures from the tissues. The sterility of the media and of the rabbit tissues used was determined with special care in all cases. The characteristics, filterability, and pathogenic powers of the bacteria isolated from the materials studied were determined according to the usual methods.

In the animal experiments young rabbits, guinea-pigs, and monkeys were used.

For microscopic work the tissues were fixed in Zenker's fluid or 10% formalin, and sections stained with hematoxylin and eosin, methylene blue and eosin, and by the Gram-Weigert and Giemsa methods.

Pieces of brain and spinal cord from some of the cases were stored in 50% glycerin in the refrigerator for future use.

Cultures were made of the blood and cerebrospinal fluid before death, and of the brain, spinal cord, spinal fluid, blood, liver, kidneys, spleen, and mesenteric lymph glands after death. In 4 cases the blood cultures and in 7 cases the cultures of the cerebrospinal fluid made before death all remained sterile. In all of these cases the material was obtained early in the course of the disease, and in some of them repeated examinations of the cerebrospinal fluid were made. Of the 10 cases of poliomyelitis studied after death 4 were from Durand Hospital of the Memorial Institute for Infectious Diseases. For the rest of the material I am indebted to Dr. John Nuzum, pathologist to Cook County Hospital, Dr. E. K. Armstrong of the health department of the city of Chicago, and Dr. E. R. LeCount, physician to the coroner of Cook County.

RESULTS OF CULTURES AND EXPERIMENTS

In 9 of the 10 cases of poliomyelitis a peculiar polymorphic streptococcus-like organism was obtained from the tissues of the central nervous system in pure or mixed culture. In 2 instances staphylococci and in 1 instance B. subtilis were associated with this organism in the brain cultures. In 5 cases the cerebrospinal fluid was examined after death and the organism isolated in pure culture in every instance. In 2 cases the liver, kidneys, spleen, and lymph nodes were cultured and this micrococcus was isolated from the mesenteric lymph nodes and kidneys in 1 case. Cultures of the Berkefeld filtrates of brain tissue from 7 cases of poliomyelitis gave the organism in 3 instances. The record of a typical case will exemplify the method of procedure and the results of this work.

Case 5.—A man, 24 years of age, entered Durand Hospital of the Memorial Institute for Infectious Diseases, Sept. 1, 1916, with what seemed like respiratory paralysis. His sickness had begun Aug. 27, 1916, with headache and general malaise. He gradually had become worse and had gone to bed August 29,

at which time fever, nausea, and vomiting were the important symptoms. On August 30, a marked weakness of the lower extremities was noticed, which gradually increased, and when he entered Washington Boulevard Hospital on August 31, he had a flaccid paralysis of both lower extremities and a weakness of the respiratory muscles. The next day the right arm and neck muscles became paralyzed. He was then transferred to Durand Hospital. The remainder of the history is unimportant for the present purpose.

The physical examination was incomplete because of the necessity for artificial respiration. Skin cold and cyanotic. Paralysis of the muscles of respiration, of both lower extremities, and of the trunk muscles. Throat, nose, and ears normal. Submaxillary glands palpable. No rigidity of the neck. Temperature 103 F. per rectum; pulse rapid, weak, and irregular. Leukocytes 15,600, 60% polymorphonuclear, 27% small mononuclear, and 12% large mononuclear. No spinal puncture made. Death September 2.

Postmortem examination was made 1 hour after death. Marked hyperemia of meninges, brain, and cord; edema of brain and cord; hyperemia of lungs, liver, kidneys, and spleen; moderate hyperplasia of mesenteric and peribronchial lymph glands; numerous small submucous hemorrhages into the stomach; calcareous tuberculosis of right and left apices; caries of the teeth.

Brain very friable, and pinkish red in color. Cerebrospinal fluid clear but increased in amount. Cord markedly hyperemic, especially in the gray substance.

Microscopically there were numerous small hemorrhages and marked round-cell perivascular infiltration into the cord, especially the gray substance; neurophagocytosis; hyperemia and edema of cord, brain, and meninges; and small gram-positive micrococci in the gray substance of the cord (Fig. 7).

Cultures of the brain, spinal cord, and spinal fluid were made September 2 in tissue ascites fluid, ascites-fluid tissue agar, ascites dextrose broth, and coagulated normal horse serum, and incubated at 35 C. under aerobic and anaerobic conditions. Bacterial growth was apparent in all the tubes, except the ascites-fluid tissue medium, after 24 hours, and a small gram-positive polymorphous micrococcus was found in pure form. After 48 hours there was a faint turbidity around the tissue in the anaerobic ascites-fluid tissue medium, which became well developed after 72 hours. In these cultures a very small gram-positive micrococcus was found in pure form. Transfers from this culture to ascites dextrose broth yielded a growth similar to that from the brain and cord in the original ascites-dextrose-broth cultures.

On September 4, a series of 6 young rabbits was injected intravenously with the washed sediment of from 6 to 8 c.c. of the original ascites-dextrose-broth cultures of the spinal cord.

Rabbit 1.—Three days after the inoculation there was a flaccid paralysis of the right foreleg. On the next day the right hindleg was paralyzed and the animal could not walk. Killed September 8. Marked hyperemia of meninges, hyperemia with edema of brain and cord, especially of the gray matter of the cord, small subpial hemorrhages into the cervical region of the cord; a few small subpleural hemorrhages; and hyperemia of kidneys and liver. Cocci similar in growth and form to the organisms injected were isolated from brain, cord, spinal fluid, and kidneys, but not from joints or heart blood. Microscopically, there were numerous small hemorrhages with round-cell perivascular infiltration, edema, and hyperemia in the gray matter of the spinal cord (Figs. 10, 11, 12, and 13); hemorrhages beneath the pia of brain and cord; and numerous cocci in the gray substance of the cord.

Rabbit 2.—Died 48 hours after inoculation without any evidences of paralysis. No gross changes. Cultures from brain, cord, cerebrospinal fluid, heart blood, and kidneys yielded cocci like those injected, in pure growth; joints sterile. Microscopically, there were numerous subpial hemorrhages; edema, hyperemia, and hemorrhages into the gray substance of the spinal cord, and cocci in the region of these hemorrhages.

Rabbit 3.—Three days after inoculation a flaccid paralysis of the right hind-leg developed. Killed on the 4th day. No gross changes other than marked hyperemia and edema of brain and cord and a small subpial hemorrhage in the lumbar region of the spinal cord. Cultures from brain and cord yielded the coccus injected, in pure growth; heart blood and joints sterile. Microscopically, there were small hemorrhages, edema, and round-cell infiltration into the gray substance of the cord; hemorrhages beneath the pia; and cocci in the region of the lesions of the cord.

Rabbit 4.—After 3 days flaccid paralysis of the right foreleg and on the 4th day, of the left foreleg and neck muscles, so that the animal could not move around. Killed. Marked hyperemia and edema of brain, cord, and meninges, and a few small submucous hemorrhages into the stomach. The small coccus was recovered from brain, cord, and cerebrospinal fluid in pure growth. Microscopically, there were hyperemia, hemorrhages, edema, and round-cell infiltration, especially in the gray substance of the cord, and hemorrhages beneath the pia of brain and cord.

Rabbits 5 and 6.—Died 24 hours after inoculation. No signs of paralysis before death. The coccus injected was recovered from the brain in Rabbit 5 and from the blood only, in Rabbit 6. No anatomic changes.

On September 11, a second series of 12 rabbits was inoculated intravenously with 24-hour ascites-dextrose-broth cultures of this organism. These cultures were made from single colonies on blood-agar plates, and the dose was the growth from 5-8 c.c. of a 24-hour ascites-dextrose-broth culture.

Seven of these animals developed flaccid paralysis of one or more extremities in from 4 to 7 days. One died 2 days after the inoculation; 5 died 4 days after the injection, only 1 of which showed paralysis. All of those living after 4 days developed paralysis in some form. In all of the 12 animals, characteristic lesions of the central nervous system were found. Grossly the changes consisted of marked hyperemia, edema of brain, spinal cord, and meninges. Microscopically, there were almost uniformly hemorrhages, round-cell infiltration, edema, and cellular degeneration especially in the gray substance of the spinal cord. In none of these animals were lesions of the joints noted, or bacteria recovered from the joints by cultural methods.

To summarize: 18 rabbits were inoculated with cultures of a streptococcus-like organism obtained from the brain and cord of a fatal case of poliomyelitis and 10 of these animals developed definite paralysis of one or more groups of muscles in from 3 to 7 days. In 15 animals marked changes, both gross and microscopic, were found which were similar to those in acute poliomyelitis in man.

In all the cases studied similar results were obtained. A polymorphic streptococcus-like organism was isolated, which, when injected into rabbits, produced acute inflammation of the central nervous sys-

tem like that in poliomyelitis in man. Similar results have been reported by Rosenow, Towne, and Wheeler[13] and Nuzum and Herzog.[14]

CHARACTERISTICS OF THE ORGANISM ISOLATED

The micrococci isolated from the brains, cords, and spinal fluids of 9 cases of poliomyelitis were in most instances similar in form and culture. On standard human-blood-agar plates (1 part of human blood to 9 parts of plain agar) the organism grew slowly, especially early after isolation. The colonies were usually somewhat dry, small, adherent, and surrounded by faint halos of greenish discoloration. After 2 or 3 days the halo became a hazy ring of hemolysis. The colonies differ greatly from those of ordinary Streptococcus viridans, pneumococcus, and Streptococcus pyogenes on blood-agar plates, but the differences are perhaps not characteristic enough for ready differentiation. In 2 instances hemolysis was noticeable in the 24-hour cultures, and in 1 instance the growth on blood agar did not appear until the 3rd day of incubation. As the cultures grew more luxuriantly on continued cultivation, the hemolytic property increased.

In ascites dextrose broth the growth of all the strains was moderate. After 18 hours there was a diffuse turbidity, which rapidly became granular with a white sediment at the side and bottom of the tube. In most instances the fluid media remained turbid. Variations in oxygen tension did not materially alter the growth of these organisms. In the anaerobic ascites-fluid tissue medium the fluid around the tissue became opalescent after from 24 to 48 hours and a fine sediment collected around the tissue, gradually a finely granular haze extended upwards, and last the whole column of fluid became turbid. As a rule, the growth on the ascites-fluid tissue medium was similar to the growth of poliomyelitis virus as described by Flexner and Noguchi.[11] Occasionally the whole column of fluid became turbid after from 24 to 48 hours and the culture resembled in some ways the aerobic ascites-dextrose-broth cultures, but usually the anaerobic cultures required from 3 to 7 days for a good growth to develop. Two strains decolorized litmus milk and liquefied gelatin. Five strains fermented inulin. None were autolyzed in bile or normal salt solution, or agglutinated by antiserum for Groups I and II of the pneumococcus.

In form, all the strains revealed similar variations. On blood agar the organisms grew as medium-sized, gram-positive, slightly oblong

[13] Jour. Am. Med. Assn., 1916, 67, p. 1202.
[14] Ibid., 1916, 67, 1205.

micrococci in pairs or short chains. In ascites dextrose broth the form was rather variable. Some were very large round or oval diplococci (Fig. 1); others medium-sized; but usually in these cultures there were variations from minute, hardly visible, bodies to large round cocci. Often chains were seen containing both extremes in size.

The cocci usually grew in pairs, short chains, or groups, and in some ascites-dextrose-broth cultures they closely resembled staphylococci. Involution forms developed in the cultures after from 7 to 14 days, both large and small forms being seen. But these involution forms tended to correspond and a characteristic change usually took place in from 10 to 14 days when the organisms became large and stained irregularly. Large cocci with clear round or oval central areas surrounded by blue-staining borders were rather numerous, and increased in number with the age of the culture (Fig. 8). These forms disappeared on inoculation into fresh media.

In the ascites-fluid tissue medium the organisms were usually very minute, ranging from 0.25 to 0.5 microns in diameter. They were gram-positive, and arranged in pairs, chains, and groups (Fig. 3), the large forms being not uncommonly intermingled with the minute forms. In this medium the organisms also showed variable involution forms as the culture became older, and the peculiar, irregularly staining bodies just described also appeared in these cultures after from 2 to 3 weeks. The tendency of the organisms was to grow in small forms in the anaerobic ascites-fluid tissue medium and in large forms in aerobic ascites-dextrose-broth cultures, but many deviations from this tendency were noted.

Cultures of this organism passed through Berkefeld filters of the "N" type in most instances, but were removed from the media by Maassen porcelain filters. The ascites-fluid tissue medium was most favorable for the growth of the filterable form of the organism, but the other culture media were not entirely unfavorable, especially ascites dextrose broth.

The organism was usually killed by a temperature of 56 C. for from 30 minutes to 1 hour, but was resistant to glycerin. A typical culture was obtained from each of 2 human poliomyelitis brains 3 months after they had been placed in 50% glycerin.

All the strains seemed not strongly virulent for rabbits, the sediment of about 5 c.c. of an ascites-dextrose-broth culture being required

TABLE 1

MORPHOLOGIC AND CULTURAL CHARACTERISTICS OF THE ORGANISM ISOLATED FROM THE CENTRAL NERVOUS SYSTEM IN ACUTE POLIOMYELITIS

Strain	Hemolysis	Ascites Dextrose Broth	Morphology	Litmus Milk	Plain Broth	Mannite
1	Small dry colonies, faint green halo, no hemolysis	Cloudy sediment	No chains, large organisms	0	—	±
3	Dry colonies, faint green halo, no hemolysis	Cloudy sediment	No chains, very large forms	0	—	±
4	Dry colonies, faint green halo, no hemolysis	Cloudy sediment	No chains, very large forms	0	—	±
5	More moist than No. 3, otherwise the same	Cloudy sediment	No chains, very large forms	0	—	±
6	Same as No. 1.....	Cloudy sediment	No chains, very large forms	0	—	±
7	Very small dry colonies, faint green halo	Cloudy sediment	No chains, very large forms	No growth	—	—
8	Small dry colonies, green halo, faint hemolysis	Cloudy sediment	Chains of 20-40 cocci, very small forms	Coagulated, discolored	—	+
9	Like 8, but with greener halo	Cloudy sediment	Chains of 10-20 cocci, very small forms	Coagulated, slightly acid	—	+
10	Small dry colonies, green halo	Cloudy sediment	Pairs and short chains, medium-sized forms	0	—	—
11*	Small dry colonies, faint green halo, hemolysis after 48 hours	Cloudy sediment	Pairs, chains, large and small forms	0	—	—

* Strain 11 was isolated from the cerebrospinal fluid obtained at autopsy from a person who had died suddenly. The pathologic changes in the brain and cord were those of acute poliomyelitis.

to cause death in from 3 to 10 days. The virulence also decreased with growth on artificial media.

In describing the poliomyelitis coccus the marked tendency to variations in form is noteworthy, especially as shown on various kinds of media. Pleomorphism, however, seems to be only a minor characteristic, representing a reaction on the part of the organism to environment. In cultures of ordinary hemolytic streptococci on almost all media just as great variations in form can be observed. Staphylococci, too, when grown on various kinds of media, appear in large and in minute forms. Streptococci and staphylococci grown under anaerobic

TABLE 1—(Continued)

MORPHOLOGIC AND CULTURAL CHARACTERISTICS OF THE ORGANISM ISOLATED FROM THE CENTRAL NERVOUS SYSTEM IN ACUTE POLIOMYELITIS

Maltose	Dextrose	Lactose	Starch	Raffinose	Salicin	Inulin	Solubility in Bile	Gelatin at 20 C.	Agglutination Test with Pneumococcus Sera	Filterability (Berkefeld N)
+	+	+	—	+	+	+	—	—	—	+
+	+	+	—	+	+	+	—	—	—	+
+	+	+	—	+	+	+	—	—	—	—
+	+	+	—	+	+	+	—	—	—	+
+	+	+	—	+	+	+	Dissolved	—	—	+
+	+	+	—	—	—	—	—	—	—	+
+	+	+	—	—	+	—	—	Liquefied	—	+
±	+	+	—	—	+	—	—	Liquefied	—	+
+	+	+	—	+	—	—	—	—	—	+
+	+	+	—	—	—	—	—	—	—	—

conditions in the ascites-fluid tissue medium over a period of from 3 to 7 days often produce changes in the medium and develop forms which resemble very closely those of the virus as described by Flexner and Noguchi. It may be said, however, that the tendency to develop peculiar forms is probably greater in the poliomyelitis coccus than in the other micrococci with which it might be confused. Again, this poliomyelitis coccus seems to produce lesions in the central nervous system, at least of rabbits, which is not the case with pneumococci, streptococci, or staphylococci as ordinarily seen.

The morphologic and cultural characteristics of the different strains studied in this work are shown in Table 1.

THE CHARACTER OF THE LESIONS PRODUCED IN RABBITS

The lesions of the central nervous system in animals, especially rabbits after intravenous, intraperitoneal, and intracerebral injection of cultures of the poliomyelitis coccus, were comparatively regular and characteristic. All methods of inoculation were successful, but the intravenous and intracerebral methods gave the most uniform results. The animals usually began to show symptoms of nervous lesions in from 3 to 7 days, but occasionally only after a much longer time. In one instance 3 weeks elapsed before the rabbit developed paralysis. The manifestations in rabbits were varied; special care was exercised to determine whether paralysis actually was present or some other condition, as arthritis, myositis, or the profound general weakness which often precedes death in these animals. In most instances the picture was striking. In from 3 to 7 days after inoculation the animal would walk with some difficulty and rapidly a flaccid paralysis of one or more limbs would develop, the general condition, however, often being still good. Paralysis of the muscles of one or more extremities, of the neck, and of the respiratory muscles was seen repeatedly. A few died early after the paralysis, sometimes in convulsions, and sometimes suddenly, with rapidly developing respiratory embarrassment.

The lesions in the rabbit were similar to those described as characteristic of acute poliomyelitis in man. Edema and hyperemia of brain and cord associated with subpial hemorrhages were almost constantly found. The brains of rabbits with nervous symptoms were always larger than normal, very moist, friable, deep-pink or red in color, with small subpial hemorrhages especially in the region of the medulla. Occasionally a large hemorrhage beneath the pia was found. The cerebrospinal fluid was turbid, increased in amount, and under pressure. And in these animals the spinal cord was swollen, moist, and often so soft that it could not be removed intact. The spinal canal was usually filled completely by the cord. On section the gray matter would often stand out as a pink area.

Microscopically, the lesions were also striking. Small subpial hemorrhages (Fig. 9) were constant, especially in the fissures of the brain and the central fissure of the cord. Hemorrhages into the gray substance (Figs. 10 and 11) of varying degrees and round-cell infiltration especially around the blood vessels (Figs. 12 and 13), with edema and hyperemia were the most noticeable changes in the cord. Neurophagocytosis, diffuse round-cell infiltration, and degeneration were frequently

present. Micrococci were demonstrated regularly in the lesions both of man (Figs. 4 and 7) and of animal (Figs. 5 and 6).

Sterile filtrates of brains of poliomyelitis patients were injected intraperitoneally into guinea-pigs and intracerebrally, intraperitoneally, and intravenously into rabbits. Doses of from 10 to 25 c.c. of 100% filtrates were used for both rabbits and guinea-pigs. In no instance did the guinea-pigs or rabbits thus inoculated show any evidences of infection.

Because of the scarcity of monkeys only a few experiments on these animals have thus far been made. Four monkeys (M. rhesus) have been inoculated with freshly isolated cultures of the poliomyelitis coccus. One received the bacteria in 15 c.c. of a 24-hour ascites-dextrose-broth culture intravenously, another a similar dose intraperitoneally, a third the growth from 0.5 c.c., and a fourth that of 1 c.c. of an ascites-dextrose-broth-culture intracerebrally. The one injected intravenously seemed ill for several days, but recovered without any manifestations of paralysis. The monkey injected intracerebrally with the organisms in 1 c.c. of a 24-hour ascites-dextrose-broth culture of a strain of the poliomyelitis coccus became ill 4 days later, and developed flaccid paralysis of the left arm. After an illness of about 2 weeks, improvement set in, and partial use of the paralyzed arm has been regained. The other two monkeys remained normal following inoculation with this organism.

Experiments bearing on the true relation of this polymorphic coccus to the virus of poliomyelitis described by Flexner and Noguchi and immunologic studies will be included in subsequent publications.

SUMMARY

In a bacteriologic examination of fresh material from 10 cases of acute poliomyelitis a peculiar polymorphic streptococcus-like organism has been isolated in 9 instances, in 7 of which the growth has been pure. Similar organisms have been demonstrated microscopically in the tissues of the central nervous system of these cases. Cultures of this coccus injected into rabbits, have produced paralysis of various groups of muscles, and characteristic lesions in the central nervous system consisting of hyperemia and edema of the tissues, with hemorrhages, round-cell perivascular infiltration, and neurophagocytosis in the spinal cord, especially in the gray substance, similar in every detail to the changes considered characteristic of acute poliomyelitis in man.

This micrococcus has been recovered from the lesions in the inoculated rabbits by both cultural and microsopic methods.

The artificial cultivation of the poliomyelitis coccus in an ascites-fluid tissue medium under anaerobic conditions causes changes in the media which cannot be differentiated from those previously described for cultures of the so-called virus of poliomyelitis. Morphologically, also, this bacterium when grown on the same media is similar to the virus, and in stained smears it appears in minute gram-positive coccus-like bodies arranged in pairs, groups, and chains. These minute forms disappear when the organism is cultivated in other media under aerobic conditions.

The morphologic, cultural, and pathogenic characters of the poliomyelitis coccus thus far determined indicate that it is an important factor in the disease.

EXPLANATION OF PLATES

PLATE 1

FIGS. 1 and 2. Large forms of the poliomyelitis coccus. Ascites-dextrose-broth culture. Gram stain. × 1200.

FIG. 3. Small forms of the poliomyelitis coccus. Anaerobic ascites-fluid tissue culture. Gram stain. × 1200.

FIG. 4. Microorganisms in stained smear of the fresh brain from a fatal case of poliomyelitis. Gram stain. × 1200.

FIGS. 5 and 6. Microorganisms in the gray substance of the spinal cord of a paralyzed rabbit dying after injection of a culture of the poliomyelitis coccus. Gram-Weigert stain. × 1200

PLATE 2

FIG. 7. Microorganisms in the gray substance of the spinal cord of a patient that died of poliomyelitis. Gram-Weigert stain. × 1200.

FIG. 8. Peculiar degeneration forms of the poliomyelitis coccus. Gram stain. × 1200.

FIG. 9. Subpial hemorrhages in the brain of a rabbit injected with the poliomyelitis coccus. Hematoxylin and eosin. × 60.

PLATE 3

FIG. 10. Small hemorrhages in the gray substance of the spinal cord of a paralyzed rabbit injected with a culture of the poliomyelitis coccus. Methylene blue and eosin. × 110.

FIG. 11. Same as Fig. 10. × 425.

PLATE 4

FIG. 12. Round-cell infiltration in the gray substance of the spinal cord of a rabbit that died after injection of a culture of the poliomyelitis coccus. Hematoxylin and eosin. × 150.

FIG. 13. Same as Fig. 12. × 425.

Plate 1

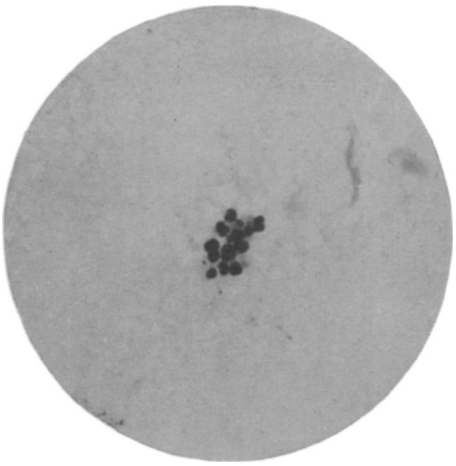

Figure 1

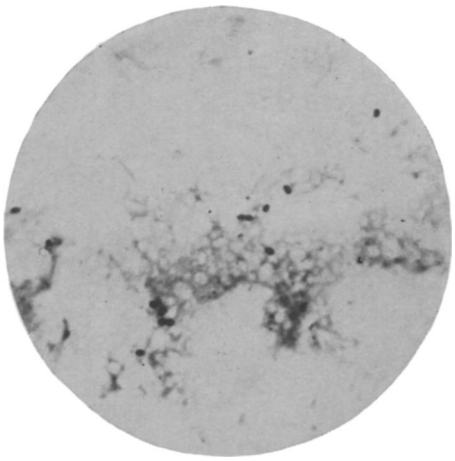

Figure 4

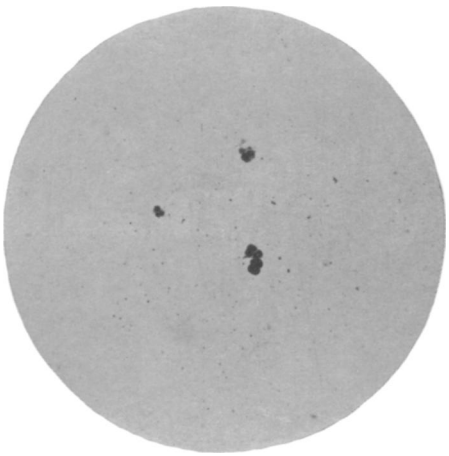

Figure 2

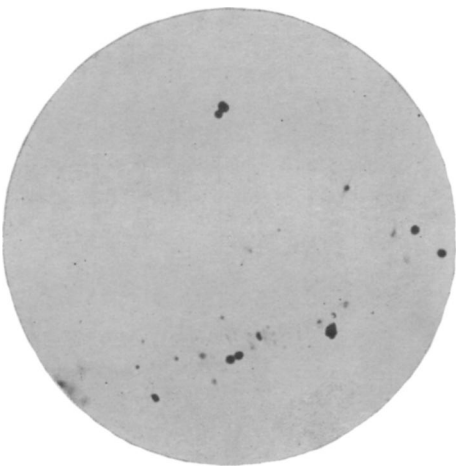

Figure 5

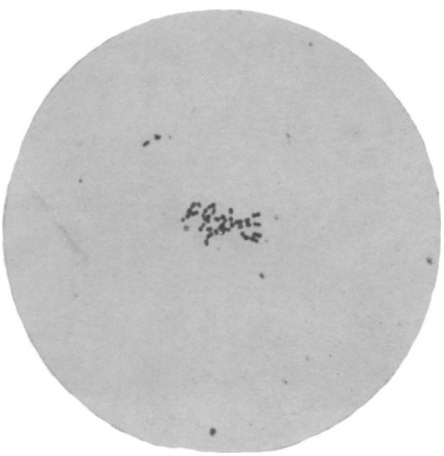

Figure 3

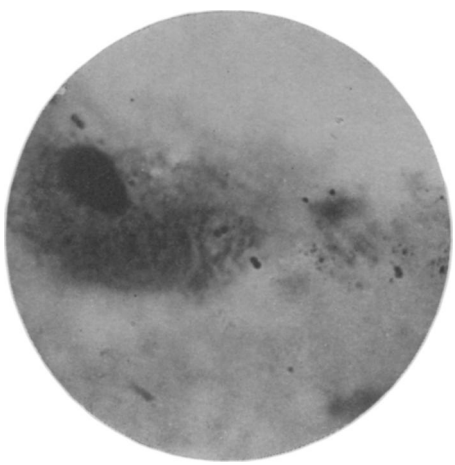

Figure 6

Plate 2

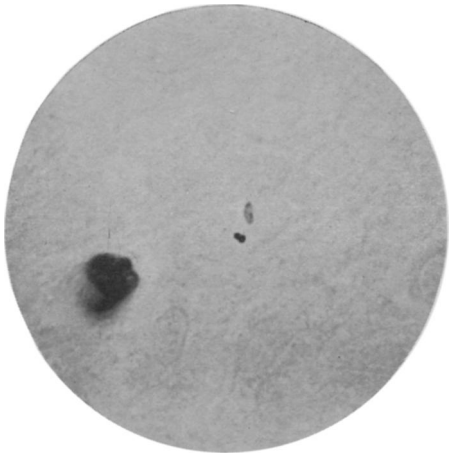

Figure 7

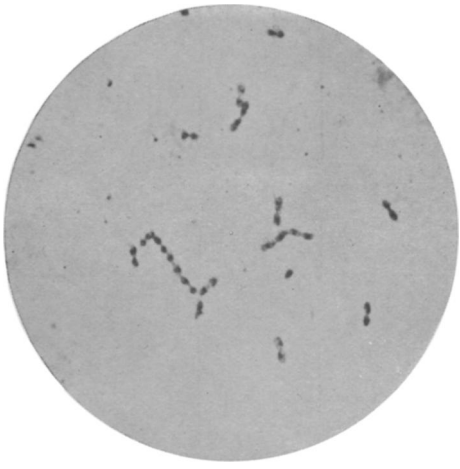

Figure 8

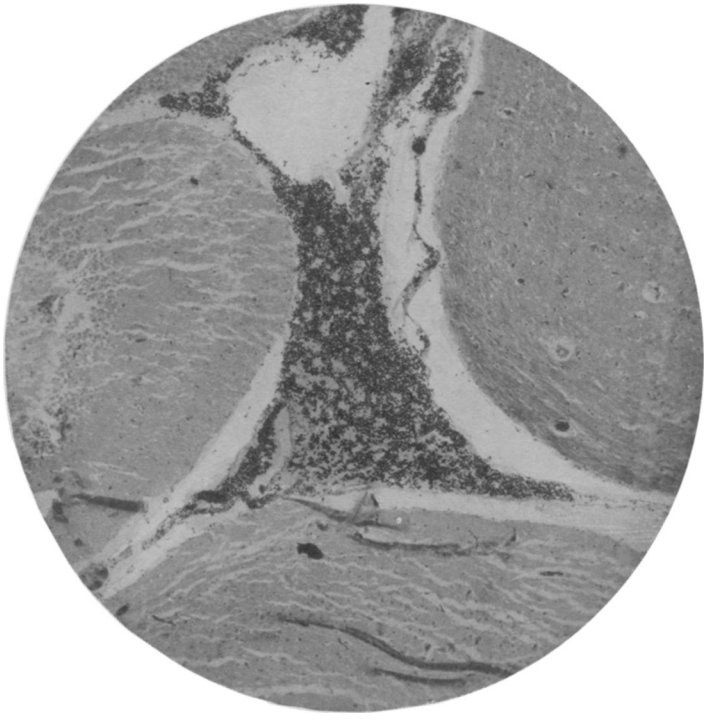

Figure 9

PLATE 3

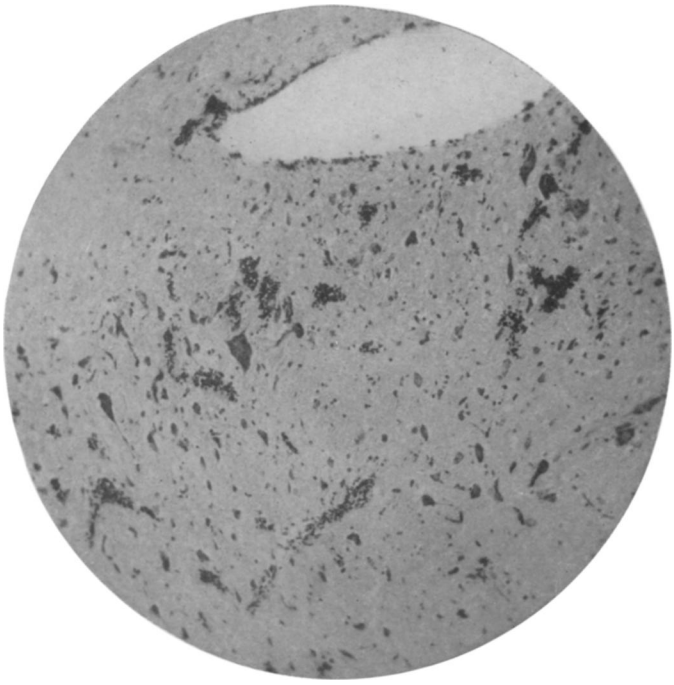

Figure 10

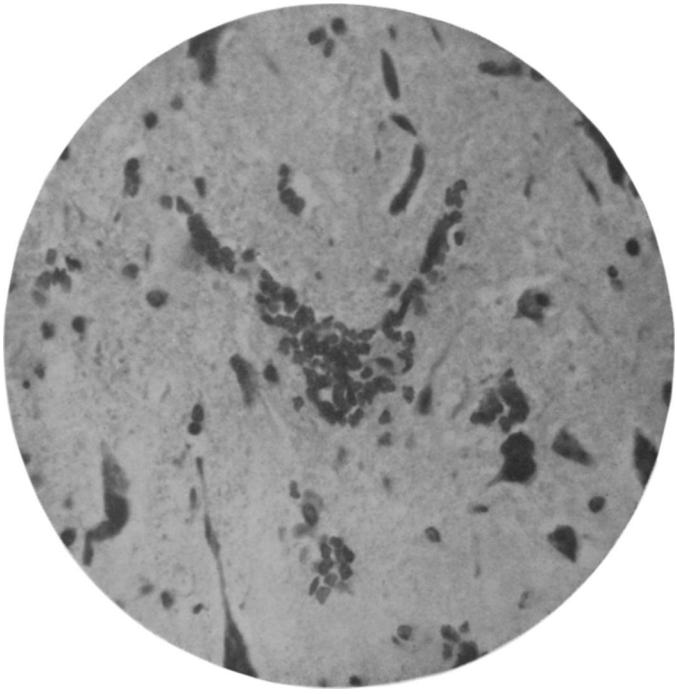

Figure 11

Plate 4

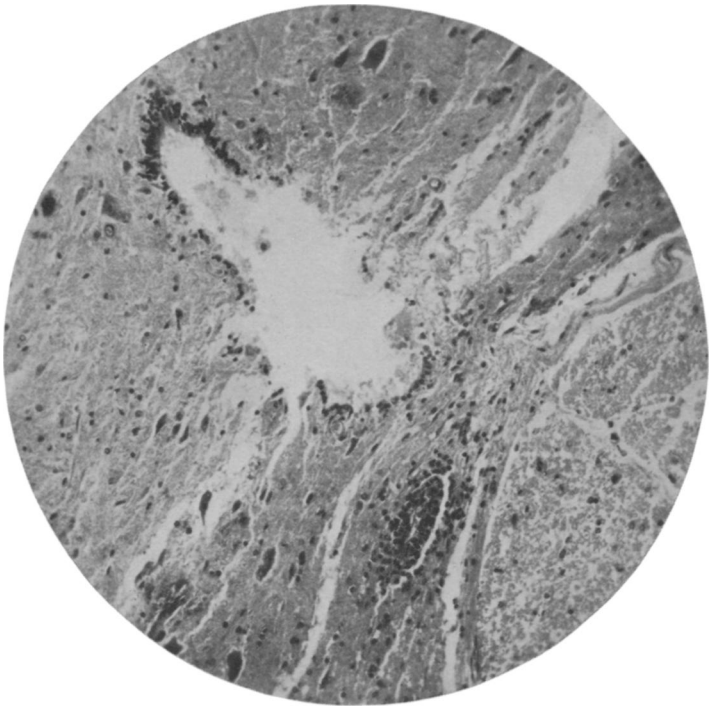

Figure 12

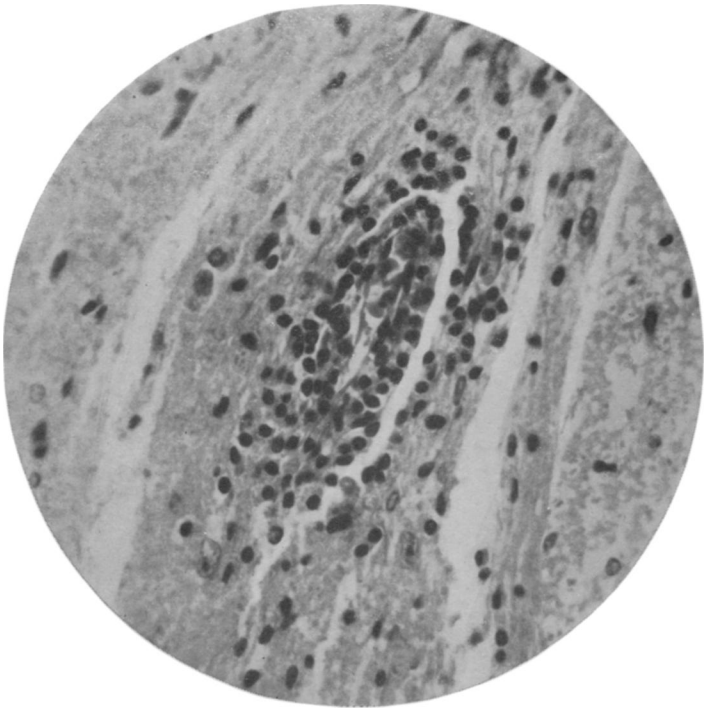

Figure 13